(N588K) in the pore forming region of the cardiac K_r channel (KCNH2, HERG). The mutations dramatically increase IK_r leading to heterogeneous abbreviation of action potential duration and refractoriness and rendering the channel relatively unresponsive to IK_r blockers. **Conclusion:** Our data point to a novel genetic mechanism responsible for sudden death in children, infants and young adults. The mutation leads to a heterogeneous abbreviation of action potential duration and refractoriness, creating the substrate for reentrant arrhythmias.

9:30 a.m.

807-2 Selective Inhibition of Human Cardiac Kir2.2 Inward Rectifier Channels by Adrenergic Alpha-1a Receptors

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Background:
Human Kir2.1 and Kir.2.2 potassium channels predominantly contribute to the molecular basis of the cardiac inwardly rectifying potassium current I_{K1}. Reduction of I_{K1} (as seen in Andersen's syndrome and in congestive heart failure) causes delayed afterdepolarizations which may result in premature ventricular beats and ventricular tachycardia. Inhibition of I_{K1} *via* activation of adrenergic alpha-1 receptors is a well-known phenomenon. However, its molecular basis has not been elucidated yet. Therefore, we investigated the interaction of alpha-1a receptors with Kir2.1 and Kir2.2 channels in the *Xenopus* oocyte expression system.
Methods:
Cloned human adrenergic alpha-1a receptors and human Kir2.1 and Kir2.2 channels were co-expressed in *Xenopus* oocytes and pharmacological experiments were performed using the double electrode voltage clamp technique.
Results:
Application of phenylephrin (10 µM) to *Xenopus* oocytes expressing only Kir2.1 or Kir2.2 channels was without any effect. However, phenylephrin (10 µM) caused a significant inhibition of Kir2.2 if the channels were co-expressed with adrenergic alpha-1a receptors. Steady state conditions were reached after 30 minutes and current amplitudes were reduced by 20 ± 5% compared to control measurements ($p<0.05$). In contrast, Kir2.1 currents were not affected by activation of the receptors.
Surprisingly, in Kir2.2 mutant channels lacking all protein kinase C (PKC) consensus sites, the same effect as in the wild type could be observed.
Conclusion:
Our study demonstrates that adrenergic alpha-1a receptors exert an inhibitory effect on human Kir2.2, but not Kir2.1 channels. The effect is independent of direct PKC mediated phosphorylation of the channel protein. These findings elucidate the molecular basis of a regulatory pathway which may contribute to the generation of ventricular arrhythmias in congestive heart failure.

9:45 a.m.

807-3 Additional Gene Modifiers Reduce Effectiveness of β-Blockers in the Long QT Type 1 Syndrome

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Background:β-blockers are widely used to prevent the lethal cardiac events that associate long QT syndrome (LQTS) especially in *KCNQ1*-related LQTS (LQT1) patients. Some LQT1 patients, however, are refractory to this therapy.
Methods and Results: Eighteen symptomatic LQTS patients (12 families) were genetically diagnosed to have heterozygous *KCNQ1* variants and received β-blocker therapy. Cardiac events recurred in 4 members (3 families) despite the continued therapy during the mean follow-up period of 70 months. Three of them (2 families) had the same mutation, A341V (*KCNQ1*), and the other had R243H (*KCNQ1*). The latter patient took aprindine that seemed to be responsible for the event. Because A341V (*KCNQ1*) has not been evaluated in heterologous mammalian expression system, we conducted functional analysis to investigate severe phenotype using COS7 cells. And we found that A341V (*KCNQ1*) is a loss-of-function type mutation (not dominant negative) as well as previous report in *Xenopus oocytes.* Further genetic screening revealed that one A341V (*KCNQ1*) family cosegregated with S706C (*KCNH2*) and another with G144S (*KCNJ2*). Produced functional outcome of S706C (*KCNH2*) mutation reduced current density with voltage shift of activation kinetics. Action potential simulation study was conducted based on the KYOTO model to estimate influences of additional gene modifiers. In both models mimicking LQT1 plus 2 and LQT1 plus 7, incidence of early afterdepolarization increased compared with LQT1 model under the setting of β adrenergic stimulation.
Conclusion: Multiple mutations in different LQTS-related genes may modify the clinical characteristics. Expanded gene survey might be required in the LQT1 patients who are resistant to β-blocker therapy.

10:00 a.m.

807-4 CUG Expansions in a Myotonic Dystrophy Mouse Model Cause Cardiac Conduction Abnormalities and Pathologic Electrophysiology Findings

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Background: Myotonic dystrophy (DM) is caused by a CTG trinucleotide expansion in the 3' untranslated region of the DM gene on chromosome 19. To assess the titration of RNA-binding proteins by expanded RNA CUG sequences on cardiac function, we studied transgenic mice carrying long and short CTG repeats, expressing the RNA CUG message at low (TG^{low}) and high (TG^{high}) levels.
Methods: 14 TG^{high}, 9 TG^{low} mice, and 6 age-matched wild-types (WT) underwent ECG recordings, in vivo electrophysiology studies, and echocardiography.
Results: ECG measurements in sedated mice demonstrated significant PR, QRS and QT prolongation in TG^{low} compared to WT mice. Right bundle branch block was seen in 16 of 22 TG versus 1 of 7 WT mice. Programmed pacing revealed inducible atrial fibrillation (2/19) and ventricular tachycardia (2/19) only in TG mice. Ventricular effective refractory period was longer in TG (TG^{low}: 63±9ms, TG^{high}: 56±13ms) than in WT mice (45±6ms, $p<0.05$). Echocardiography showed hypertrophic appearance only in TG^{high} mice, with a shorter end diastolic diameter and thicker left ventricular posterior wall diameter ($p<0.05$).
Conclusion: *Transgenic mice overexpressing CUG repeats displayed 1^{o} heart block, infraHisian conduction abnormalities, and arrhythmia vulnerability. These features were more prominent in TG^{low} compared to TG^{high} mice. Our study supports the hypothesis that larger CTG expansions are associated with a higher risk of cardiac conduction disease in DM.*

ECG measurements in sedated mice

	WT	TG^{low}	TG^{high}	*One way ANOVA, p=*	*Post hoc Scheffe's analysis*
SCL (ms)	131±6	142±7	143±19	*ns*	*ns*
HR (bpm)	457±22	424±20	427±56	*ns*	*ns*
PR (ms)	35.0±2.8*	41.2±5.5*	37.3±3.4	*0.02*	**: p=0.03*
QRS (ms)	14.0±2.7	16.8±1.8*	13.7±2.5*	*0.01*	**: p=0.02*
QT (ms)	21.7±2.1*	30.0±2.9*°	22.6±3.7°	*0.000*	**, °: p=0.000*
QTc (ms)	19.0±2.2*	25.2±2.5*°	19.1±4.0°	*0.000*	**, °: p=0.000*

10:15 a.m.

807-5 Extracellular pH Modifies Drug Blockade on the Human Ether-A-Go-Go Related Gene (HERG) Channels

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Background: HERG encodes the rapid component of delayed rectifier potassium current (IKr), which is critical for the repolarization of cardiac action potentials. Significant change in pH can occur during ischemia and reperfusion, and the effects of pH on IKr could be an important underlying mechanism for arrhythmias.
Methods: We investigated the effect of external pH on IKr, as well as the influence of external pH on the effect of IKr inhibition by anti-arrhythmia agents. HERG expressed in Xenopus oocytes was used as the experimental model.
Results: Extracellular acidification decreased the amplitudes of both relatively small outward currents and the large tail outward currents, while the decay of tail current was accelerated. The steady state current was decreased by 14.4±6.8%, 19.8±10.4%, 23.8±10.4%, 28.3±10.5%, and the tail current was decreased by 6.7±1.1%, 8.7±2.1%, 13.4±1.5%, and 26.4±3.0% when decreasing pH from 8.0 to 7.4, 7.0, 6.6, and 6.2, respectively. The effect of acidification and IKr inhibition by quinidine, dofetilide, and azimilide was evaluated. There was no additive or synergistic effect between extracellular pH and antiarrhythmics on HERG. Quinidine 10 µM inhibited HERG tail current by 37.2±4.8% at pH 7.4. This inhibition was increased to 50.9±4.7% when extracellular pH was 8.0 and decreased to 4.5±1.8% when extracellular pH was 6.2. Dofetilide 0.3 µM inhibited HERG tail current by 64.4±7.4, 33.5±2.6 and 1.4±2.4% at extracellular pH 8.0, 7.4, and 6.2 respectively. Azimilide 10 µM inhibited HERG tail current by 63.0±3.6, 58.7±2.9 and 17.4±3.2% at extracellular pH 8.0, 7.4, and 6.2 respectively. For quinidine, dofetilide, and azimilide, one way ANOVA test showed that there was a significant difference in current block among the different pH groups.
Conclusion: Our data suggests that extracellular acidification reduces HERG current. The effects of quinidine, dofetilide, and azimilide are decreased at a more acid pH and the block of IKr enhanced at a more alkaline pH.